

# SUMMARY OF THE INVENTION

Throughout this specification, unless the context requires otherwise, the word “comprise,” or variations such as “comprises” or “comprising,” will be understood to imply the inclusion of a stated element or integer or group of elements or integers but not the exclusion of any other element or integer or group of elements or integers.

One aspect of the present invention contemplates an HBV variant exhibiting a replication fitness, in the presence of a nucleoside analogue, similar to or greater than in the absence of said nucleoside analogue.

Another aspect of the present invention provides an HBV variant carrying a mutation in the nucleoside sequence encoding a DNA polymerase resulting in an amino acid addition, substitution and/or deletion in said DNA polymerase in one or more amino acids as set forth in Formulae I and/or II:

## FORMULA I

L, B<sub>1</sub>, B<sub>2</sub>, D, W, G, P, C, B<sub>3</sub>, B<sub>4</sub>, H, G, B<sub>5</sub>, H, B<sub>6</sub>, I, R, B<sub>7</sub>, P, R, T, P, B<sub>8</sub>, R, V, B<sub>9</sub>, G, G, V, F, L, V, D, K, N, P, H, N, T, B<sub>10</sub>, E, S, B<sub>11</sub>, L, B<sub>12</sub>, V, D, F, S, Q, F, S, R, G, B<sub>13</sub>, B<sub>14</sub>, B<sub>15</sub>, V, S, W, P, K, F, A, V, P, N, L, B<sub>16</sub>, S, L, T, N, L, L, S\* (SEQ ID NO:1)

wherein:

- B<sub>1</sub> is L, or R, or I
- B<sub>2</sub> is E, or D
- B<sub>3</sub> is T, or D, or A, or N, or Y
- B<sub>4</sub> is E, or D
- B<sub>5</sub> is E, or K, or Q
- B<sub>6</sub> is H, or R, or N,
- B<sub>7</sub> is I, or T
- B<sub>8</sub> is A, or S
- B<sub>9</sub> is T or R
- B<sub>10</sub> is A, or T, or S
- B<sub>11</sub> is R, or T

- B<sub>12</sub> is V, or G  
B<sub>13</sub> is S, or I, or T, or N, or V  
B<sub>14</sub> is T, or S, or H, or Y  
B<sub>15</sub> is R, or H, or K, or Q  
5 B<sub>16</sub> is Q, or P;

and

### FORMULA II

S Z<sub>1</sub> L S W L S L D V S A A F Y H Z<sub>2</sub> P L H P A A M P H L L Z<sub>3</sub> G S S G L Z<sub>4</sub> R Y V A R  
10 L S S Z<sub>5</sub> S Z<sub>6</sub> Z<sub>7</sub> X N Z<sub>8</sub> Q Z<sub>9</sub> Z<sub>10</sub> X X X Z<sub>11</sub> L H Z<sub>12</sub> Z<sub>13</sub> C S R Z<sub>14</sub> L Y V S L Z<sub>15</sub> L L Y Z<sub>16</sub>  
T Z<sub>17</sub> G Z<sub>18</sub> K L H L Z<sub>19</sub> Z<sub>20</sub> H P I Z<sub>21</sub> L G F R K Z<sub>22</sub> P M G Z<sub>23</sub> G L S P F L L A Q F T S A I  
Z<sub>24</sub> Z<sub>25</sub> Z<sub>26</sub> Z<sub>27</sub> Z<sub>28</sub> R A F Z<sub>29</sub> H C Z<sub>30</sub> Z<sub>31</sub> F Z<sub>32</sub> Y M<sup>\*</sup> D D Z<sub>33</sub> V L G A Z<sub>34</sub> Z<sub>35</sub> Z<sub>36</sub> Z<sub>37</sub> H Z<sub>38</sub>  
E Z<sub>39</sub> L Z<sub>40</sub> Z<sub>41</sub> Z<sub>42</sub> Z<sub>43</sub> Z<sub>44</sub> Z<sub>45</sub> Z<sub>46</sub> L L Z<sub>47</sub> Z<sub>48</sub> G I H L N P Z<sub>49</sub> K T K R W G Y S L N F M G  
Y Z<sub>50</sub> I G (SEQ ID NO:2)

15

wherein:

- X is any amino acid;  
Z<sub>1</sub> is N or D;  
20 Z<sub>2</sub> is I or P;  
Z<sub>3</sub> is I or V;  
Z<sub>4</sub> is S or D;  
Z<sub>5</sub> is T or N;  
Z<sub>6</sub> is R or N;  
25 Z<sub>7</sub> is N or I;  
Z<sub>8</sub> is N or Y or H;  
Z<sub>9</sub> is H or Y;  
Z<sub>10</sub> is G or R;  
Z<sub>11</sub> is D or N;  
30 Z<sub>12</sub> is D or N;  
Z<sub>13</sub> is S or Y;  
Z<sub>14</sub> is N or Q;

Z<sub>15</sub> is L or M;  
Z<sub>16</sub> is K or Q;  
Z<sub>17</sub> is Y or F;  
Z<sub>18</sub> is R or W;  
5 Z<sub>19</sub> is Y or L;  
Z<sub>20</sub> is S or A;  
Z<sub>21</sub> is I or V;  
Z<sub>22</sub> is I or L;  
Z<sub>23</sub> is V or G;  
10 Z<sub>24</sub> is C or L;  
Z<sub>25</sub> is A or S;  
Z<sub>26</sub> is V or M;  
Z<sub>27</sub> is V or T;  
Z<sub>28</sub> is R or C;  
15 Z<sub>29</sub> is F or P;  
Z<sub>30</sub> is L or V;  
Z<sub>31</sub> is A or V;  
Z<sub>32</sub> is S or A;  
Z<sub>33</sub> is V or L or M;  
20 Z<sub>34</sub> is K or R;  
Z<sub>35</sub> is S or T;  
Z<sub>36</sub> is V or G;  
Z<sub>37</sub> is Q or E;  
Z<sub>38</sub> is L or S or R;  
25 Z<sub>39</sub> is S or F;  
Z<sub>40</sub> is F or Y;  
Z<sub>41</sub> is T or A;  
Z<sub>42</sub> is A or S;  
Z<sub>43</sub> is V or I;  
30 Z<sub>44</sub> is T or C;  
Z<sub>45</sub> is N or S;  
Z<sub>46</sub> is F or V;

- Z<sub>47</sub> is S or D;  
Z<sub>48</sub> is L or V;  
Z<sub>49</sub> is N or Q;  
Z<sub>50</sub> is V or I; and  
5 M\* is amino acid 550

and wherein S\* in Formula I is designated as amino acid 420 and the first S in Formula II is designated as amino acid 421;

and wherein said variant exhibits a replication fitness in the presence of a nucleoside analogue  
10 similar to or greater than in the absence of said nucleoside analogue.

Yet another aspect of the present invention is directed to an HBV variant comprising a mutation in the nucleotide sequence encoding the HBV surface antigen which results in an amino acid addition, substitution and/or deletion in said surface antigen in a region  
15 corresponding to the amino acid sequences set forth in Formulae I and/or II wherein said variant exhibits a replication fitness in the presence of a nucleoside analogue similar to or greater than in the absence of said nucleoside analogue.

Even yet another aspect of the present invention is directed to an HBV variant comprising a mutation in the nucleotide sequence encoding the HBV surface antigen which  
20 results in an amino acid addition, substitution and/or deletion in said surface antigen in a region corresponding to the amino acid sequences set forth in Formulae I and/or II wherein said variant results in HBV DNA levels in the presence of a nucleoside analogue similar to or greater than the levels detected in pretreated patients.

Still yet another aspect of the present invention provides an HBV comprising a  
25 mutation in the nucleotide sequences encoding a DNA polymerase and a mutation in the nucleotide sequences encoding the surface antigen wherein each mutation results in an amino acid addition, substitution and/or deletion to each of the DNA polymerase and surface antigen and wherein said variants exhibits a replication fitness in the presence of a nucleoside analogue similar to or greater than in the absence of said nucleoside analogue.

30 Another aspect of the present invention contemplates a method for determining whether an HBV strain exhibits reduced sensitivity to a nucleoside analogue, said method comprising isolating DNA or corresponding mRNA from said HBV and screening for a

mutation in the nucleotide sequence encoding the DNA polymerase and optionally the surface antigen (listed below in parenthesis) wherein the presence of a T474N (P120T), M550V (I195M), M550I (W196S), L526M, W499S/W499Q (G145R) mutation, or combinations thereof or an equivalent one or more other mutation is indicative of a variant wherein said  
5 variant exhibits a replication fitness in the presence of a nucleoside analogue similar to or greater than in the absence of said nucleoside analogue.

Another aspect of the present invention contemplates a method for detecting an HBV agent which exhibits inhibitory activity to an HBV, said method comprising:

10 generating a genetic construct comprising a replication competent-effective amount of the genome from said HBV contained in a plasmid vector and then transfecting said cells with said construct;  
contacting said cells, before, during and/or after transfection, with the agent to be tested;  
15 culturing said cells for a time and under conditions sufficient for the HBV to replicate, express genetic sequences and/or assemble and/or release virus or virus-like particles if resistant to said agent; and  
subjecting the cells, cell lysates or culture supernatant fluid to viral- or viral-component-detection means to determine whether or not the virus has  
20 replicated, expressed genetic material and/or assembled and/or been released in the presence of said agent.

Still another aspect of the present invention provides a method for detecting an HBV agent which exhibits inhibitory activity to an HBV, said method comprising:

25 generating a genetic construct comprising a replication competent-effective amount of the genome from said HBV contained in or fused to an amount of a baculovirus genome effective to infect cells and then infecting said cells with said construct;  
contacting said cells, before, during and/or after infection, with the agent to be tested;  
30 culturing said cells for a time and under conditions sufficient for the HBV to replicate, express genetic sequences and/or assemble and/or release virus or virus-like particles if resistant to said agent; and

**FIG. 10** is a diagrammatic representation of a cross-section of a magnetic storage medium.

**FIG. 11** is a diagrammatic representation of a cross-section of an optically readable data storage system.

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#### **ABBREVIATIONS**

<b>ABBREVIATION</b>	<b>DESCRIPTION</b>
LAM	lamivudine
3TC	(LAM); (-)- $\beta$ -2'-deoxy-3'-thiacytidine
HBIG	Hepatitis B immunoglobulin
HBV	Hepatitis B virus
ER	Endoplasmic reticulum

#### **DESCRIPTION OF ILLUSTRATIVE EMBODIMENTS**

Lamivudine (LAM or 3TC) is a potent inhibitor of HBV replication. It is observed that  
10 HBV DNA titres are reduced in the sera of chronically infected patients after OLT and  
treatment with LAM and HBIG. LAM inhibits viral DNA synthesis. However, after a few  
months, there is an increase in HBV titres. Levels rose to pre-treatment levels. In accordance  
with the present invention, the inventors sequenced the genomes of the HBV resistant variants  
and revealed a number of mutations in the HBV polymerase gene which resulted in a level of  
15 replication fitness in the presence of a nucleoside analogue relative to its absence. Such a  
phenomenon is demonstrable by detecting viral load or burden in patients exposed to the  
nucleoside analogue. Viral load or burden is conveniently determined by detecting viral  
nucleic acid molecules (*e.g.*, DNA), replicative intermediates, polymerase activity, levels of  
surface antigen and/or titre of viral particles. The detection of such replication fit HBV  
20 variants in the presence of a nucleoside analogue is an important step in determining an  
appropriate therapeutic protocol for patients.

Accordingly, one aspect of the present invention contemplates an HBV variant  
exhibiting a replication fitness in the presence of a nucleoside analogue similar to or greater  
than in the absence of said nucleoside analogue.

25 Preferably, the HBV variant carries a mutation in the nucleotide sequence encoding the  
HBV DNA polymerase. Such mutation results in an addition, substitution and/or deletion of

an amino acid sequence of the DNA polymerase. Reference to the HBV DNA polymerase includes domains F and A through E set forth in Formula I below:

# **FORMULA I**

5

L, B<sub>1</sub>, B<sub>2</sub>, D, W, G, P, C, B<sub>3</sub>, B<sub>4</sub>, H, G, B<sub>5</sub>, H, B<sub>6</sub>, I, R, B<sub>7</sub>, P, R, T, P, B<sub>8</sub>, R, V, B<sub>9</sub>, G, G, V, F, L, V, D, K, N, P, H, N, T, B<sub>10</sub>, E, S, B<sub>11</sub>, L, B<sub>12</sub>, V, D, F, S, Q, F, S, R, G, B<sub>13</sub>, B<sub>14</sub>, B<sub>15</sub>, V, S, W, P, K, F, A, V, P, N, L, B<sub>16</sub>, S, L, T, N, L, L, S\* (SEQ ID NO:1)

10 wherein:

B<sub>1</sub> is L, or R, or I

B<sub>2</sub> is E, or D

B<sub>3</sub> is T, or D, or A, or N, or Y

B<sub>4</sub> is E, or D

15 B<sub>5</sub> is E, or K, or Q

B<sub>6</sub> is H, or R, or N,

B<sub>7</sub> is I, or T

B<sub>8</sub> is A, or S

B<sub>9</sub> is T or R

20 B<sub>10</sub> is A, or T, or S

B<sub>11</sub> is R, or T

B<sub>12</sub> is V, or G

B<sub>13</sub> is S, or I, or T, or N, or V

B<sub>14</sub> is T, or S, or H, or Y

25 B<sub>15</sub> is R, or H, or K, or Q

B<sub>16</sub> is Q, or P;

and wherein S\* is designated as amino acid 420.

30 In this specification, reference is particularly made to the conserved regions as defined by Poch *et al.* (16) as domains A to E (see also reference 17). Regions A to E are defined by the amino acid sequence set forth in Formula II below:

# FORMULA II

S Z<sub>1</sub> L S W L S L D V S A A F Y H Z<sub>2</sub> P L H P A A M P H L L Z<sub>3</sub> G S S G L Z<sub>4</sub> R Y V A R  
L S S Z<sub>5</sub> S Z<sub>6</sub> Z<sub>7</sub> X N Z<sub>8</sub> Q Z<sub>9</sub> Z<sub>10</sub> X X X Z<sub>11</sub> L H Z<sub>12</sub> Z<sub>13</sub> C S R Z<sub>14</sub> L Y V S L Z<sub>15</sub> L L Y Z<sub>16</sub>  
5 T Z<sub>17</sub> G Z<sub>18</sub> K L H L Z<sub>19</sub> Z<sub>20</sub> H P I Z<sub>21</sub> L G F R K Z<sub>22</sub> P M G Z<sub>23</sub> G L S P F L L A Q F T S A I  
Z<sub>24</sub> Z<sub>25</sub> Z<sub>26</sub> Z<sub>27</sub> Z<sub>28</sub> R A F Z<sub>29</sub> H C Z<sub>30</sub> Z<sub>31</sub> F Z<sub>32</sub> Y M<sup>\*</sup> D D Z<sub>33</sub> V L G A Z<sub>34</sub> Z<sub>35</sub> Z<sub>36</sub> Z<sub>37</sub> H Z<sub>38</sub>  
E Z<sub>39</sub> L Z<sub>40</sub> Z<sub>41</sub> Z<sub>42</sub> Z<sub>43</sub> Z<sub>44</sub> Z<sub>45</sub> Z<sub>46</sub> L L Z<sub>47</sub> Z<sub>48</sub> G I H L N P Z<sub>49</sub> K T K R W G Y S L N F M G  
Y Z<sub>50</sub> I G (SEQ ID NO:2)

10 wherein:

- X is any amino acid;
- Z<sub>1</sub> is N or D;
- Z<sub>2</sub> is I or P;
- 15 Z<sub>3</sub> is I or V;
- Z<sub>4</sub> is S or D;
- Z<sub>5</sub> is T or N;
- Z<sub>6</sub> is R or N;
- Z<sub>7</sub> is N or I;
- 20 Z<sub>8</sub> is N or Y or H;
- Z<sub>9</sub> is H or Y;
- Z<sub>10</sub> is G or R;
- Z<sub>11</sub> is D or N;
- Z<sub>12</sub> is D or N;
- 25 Z<sub>13</sub> is S or Y;
- Z<sub>14</sub> is N or Q;
- Z<sub>15</sub> is L or M;
- Z<sub>16</sub> is K or Q;
- Z<sub>17</sub> is Y or F;
- 30 Z<sub>18</sub> is R or W;
- Z<sub>19</sub> is Y or L;
- Z<sub>20</sub> is S or A;



- $Z_{21}$  is I or V;  
 $Z_{22}$  is I or L;  
 $Z_{23}$  is V or G;  
 $Z_{24}$  is C or L;  
5  $Z_{25}$  is A or S;  
 $Z_{26}$  is V or M;  
 $Z_{27}$  is V or T;  
 $Z_{28}$  is R or C;  
 $Z_{29}$  is F or P;  
10  $Z_{30}$  is L or V;  
 $Z_{31}$  is A or V;  
 $Z_{32}$  is S or A;  
 $Z_{33}$  is V or L or M;  
 $Z_{34}$  is K or R;  
15  $Z_{35}$  is S or T;  
 $Z_{36}$  is V or G;  
 $Z_{37}$  is Q or E;  
 $Z_{38}$  is L or S or R;  
 $Z_{39}$  is S or F;  
20  $Z_{40}$  is F or Y;  
 $Z_{41}$  is T or A;  
 $Z_{42}$  is A or S;  
 $Z_{43}$  is V or I;  
 $Z_{44}$  is T or C;  
25  $Z_{45}$  is N or S;  
 $Z_{46}$  is F or V;  
 $Z_{47}$  is S or D;  
 $Z_{48}$  is L or V;  
 $Z_{49}$  is N or Q;  
30  $Z_{50}$  is V or I; and  
 $M^*$  is amino acid 550

and wherein the first S is designated as amino acid 421.

According, another aspect of the present invention provides an HBV variant carrying a mutation in the nucleoside sequence encoding a DNA polymerase resulting in an amino acid addition, substitution and/or deletion in said DNA polymerase in one or more amino acids as set forth in Formulae I and/or II:

### FORMULA I

10 L, B<sub>1</sub>, B<sub>2</sub>, D, W, G, P, C, B<sub>3</sub>, B<sub>4</sub>, H, G, B<sub>5</sub>, H, B<sub>6</sub>, I, R, B<sub>7</sub>, P, R, T, P, B<sub>8</sub>, R, V, B<sub>9</sub>, G, G, V, F, L, V, D, K, N, P, H, N, T, B<sub>10</sub>, E, S, B<sub>11</sub>, L, B<sub>12</sub>, V, D, F, S, Q, F, S, R, G, B<sub>13</sub>, B<sub>14</sub>, B<sub>15</sub>, V, S, W, P, K, F, A, V, P, N, L, B<sub>16</sub>, S, L, T, N, L, L, S\* (SEQ ID NO:1)

wherein:

15

B<sub>1</sub> is L, or R, or I

B<sub>2</sub> is E, or D

B<sub>3</sub> is T, or D, or A, or N, or Y

B<sub>4</sub> is E, or D

20 B<sub>5</sub> is E, or K, or Q

B<sub>6</sub> is H, or R, or N,

B<sub>7</sub> is I, or T

B<sub>8</sub> is A, or S

B<sub>9</sub> is T or R

25 B<sub>10</sub> is A, or T, or S

B<sub>11</sub> is R, or T

B<sub>12</sub> is V, or G

B<sub>13</sub> is S, or I, or T, or N, or V

B<sub>14</sub> is T, or S, or H, or Y

30 B<sub>15</sub> is R, or H, or K, or Q

B<sub>16</sub> is Q, or P;

and

## FORMULA II

5    S Z<sub>1</sub> L S W L S L D V S A A F Y H Z<sub>2</sub> P L H P A A M P H L L Z<sub>3</sub> G S S G L Z<sub>4</sub> R Y V A R  
      L S S Z<sub>5</sub> S Z<sub>6</sub> Z<sub>7</sub> X N Z<sub>8</sub> Q Z<sub>9</sub> Z<sub>10</sub> X X X Z<sub>11</sub> L H Z<sub>12</sub> Z<sub>13</sub> C S R Z<sub>14</sub> L Y V S L Z<sub>15</sub> L L Y Z<sub>16</sub>  
      T Z<sub>17</sub> G Z<sub>18</sub> K L H L Z<sub>19</sub> Z<sub>20</sub> H P I Z<sub>21</sub> L G F R K Z<sub>22</sub> P M G Z<sub>23</sub> G L S P F L L A Q F T S A I  
      Z<sub>24</sub> Z<sub>25</sub> Z<sub>26</sub> Z<sub>27</sub> Z<sub>28</sub> R A F Z<sub>29</sub> H C Z<sub>30</sub> Z<sub>31</sub> F Z<sub>32</sub> Y M<sup>\*</sup> D D Z<sub>33</sub> V L G A Z<sub>34</sub> Z<sub>35</sub> Z<sub>36</sub> Z<sub>37</sub> H Z<sub>38</sub>  
      E Z<sub>39</sub> L Z<sub>40</sub> Z<sub>41</sub> Z<sub>42</sub> Z<sub>43</sub> Z<sub>44</sub> Z<sub>45</sub> Z<sub>46</sub> L L Z<sub>47</sub> Z<sub>48</sub> G I H L N P Z<sub>49</sub> K T K R W G Y S L N F M G  
      Y Z<sub>50</sub> I G (SEQ ID NO:2)

10

wherein:

- X     is any amino acid;
- Z<sub>1</sub>    is N or D;
- 15    Z<sub>2</sub>    is I or P;
- Z<sub>3</sub>    is I or V;
- Z<sub>4</sub>    is S or D;
- Z<sub>5</sub>    is T or N;
- Z<sub>6</sub>    is R or N;
- 20    Z<sub>7</sub>    is N or I;
- Z<sub>8</sub>    is N or Y or H;
- Z<sub>9</sub>    is H or Y;
- Z<sub>10</sub>    is G or R;
- Z<sub>11</sub>    is D or N;
- 25    Z<sub>12</sub>    is D or N;
- Z<sub>13</sub>    is S or Y;
- Z<sub>14</sub>    is N or Q;
- Z<sub>15</sub>    is L or M;
- Z<sub>16</sub>    is K or Q;
- 30    Z<sub>17</sub>    is Y or F;
- Z<sub>18</sub>    is R or W;
- Z<sub>19</sub>    is Y or L;

$Z_{20}$  is S or A;  
 $Z_{21}$  is I or V;  
 $Z_{22}$  is I or L;  
 $Z_{23}$  is V or G;  
5  $Z_{24}$  is C or L;  
 $Z_{25}$  is A or S;  
 $Z_{26}$  is V or M;  
 $Z_{27}$  is V or T;  
 $Z_{28}$  is R or C;  
10  $Z_{29}$  is F or P;  
 $Z_{30}$  is L or V;  
 $Z_{31}$  is A or V;  
 $Z_{32}$  is S or A;  
 $Z_{33}$  is V or L or M;  
15  $Z_{34}$  is K or R;  
 $Z_{35}$  is S or T;  
 $Z_{36}$  is V or G;  
 $Z_{37}$  is Q or E;  
 $Z_{38}$  is L or S or R;  
20  $Z_{39}$  is S or F;  
 $Z_{40}$  is F or Y;  
 $Z_{41}$  is T or A;  
 $Z_{42}$  is A or S;  
 $Z_{43}$  is V or I;  
25  $Z_{44}$  is T or C;  
 $Z_{45}$  is N or S;  
 $Z_{46}$  is F or V;  
 $Z_{47}$  is S or D;  
 $Z_{48}$  is L or V;  
30  $Z_{49}$  is N or Q;  
 $Z_{50}$  is V or I; and  
 $M^*$  is amino acid 550

and wherein S\* in Formula I is designated as amino acid 420 and the first S in Formula II is designated as amino acid 421;

and wherein said variant exhibits a replication fitness in the presence of a nucleoside analogue  
5 similar to or greater than in the absence of said nucleoside analogue.

Preferred nucleoside analogues, including FAM and/or LAM and their chemical derivatives and homologues, are those which select mutations in the B and/or C domains of HBV polymerase.

10 Furthermore, in one particular embodiment, the nucleoside analogue selects a corresponding mutation in the HBV surface antigen gene resulting in an HBIG-resistant mutant. In another particularly preferred embodiment, the replication fitness HBV variant is selected following exposure to both the nucleoside analogue and HBIG treatment.

Accordingly, another aspect of the present invention is directed to an HBV variant  
15 comprising a mutation in the nucleotide sequence encoding the HBV surface antigen which results in an amino acid addition, substitution and/or deletion in said surface antigen in a region corresponding to the amino acid sequences set forth in Formulae I and/or II wherein said variants exhibits a replication fitness in the presence of a nucleoside analogue similar to or greater than in the absence of said nucleoside analogue.

20 In a related embodiment of the present invention, there is provided an HBV variant comprising a mutation in the nucleotide sequence encoding the HBV surface antigen which results in an amino acid addition, substitution and/or deletion in said surface antigen in a region corresponding to the amino acid sequences set forth in Formulae I and/or II wherein said variant results in HBV DNA levels in the presence of a nucleoside analogue similar to or  
25 greater than the levels detected in pretreated patients.

More particularly, the present invention provides an HBV comprising a mutation in the nucleotide sequences encoding a DNA polymerase and a mutation in the nucleotide sequences encoding the surface antigen wherein each mutation results in an amino acid addition, substitution and/or deletion to each of the DNA polymerase and surface antigen and wherein  
30 said variants exhibits a replication fitness in the presence of a nucleoside analogue similar to or greater than in the absence of said nucleoside analogue.